VarIso1

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VarIso1 is a PC-compatible program that uses empirical diet and $\delta^{13}C$ stable isotope data to generate an expected relationship between diet and isotope variation.

Downloading this program will place a single .exe file in the folder you save it to.

Overview of the program:

Double-clicking the icon for the program will open an MS-DOS window, and the program will start. You will go through the following steps (discussed in detail further on in this manual):

1. You will be prompted to enter the number of individuals in your sample.
2. You will be prompted to enter the number of prey categories.
3. You will be prompted to enter the vector of proportions of the population overall diet.
4. You will be prompted to enter the vector of prey isotopic signatures.
5. You will be asked if you want to enter the prey dry masses.
    a. If you answer 'yes', you will be prompted to enter the vector of prey dry weights.
6. You will be prompted to enter the number of replicate simulations at a given level of diet variation.

Program details:

The idea underlying our simulations is to generate a high number of simulated populations using a set of parameters that you obtained from your empirical sample. For each simulated population, a standardized index of among-individual diet variation and the isotopic variance will be calculated. With these measures you will be able to establish an expected relationship between among-individual diet variation and isotopic variance,

which in turn will allow the conversion of the empirical isotopic variance into a measure of diet variation that can be compared across different populations. Details on how this can be done are given below.

Heading numbers follow the preceding outline of the program.

1) Number of individuals

Here you should enter the number of individuals in your empirical sample. In the simulations, populations of this size will be generated (see below).

2) Number of prey categories

Here you should enter the number of prey categories identified in your empirical sample. In the simulations, individuals will be allowed to feed on the number of prey categories you enter (see below).

3) Population diet

Here you should enter the population diet vector **p** containing the proportions q_j of the j resources in the population diet, so that $\mathbf{p} = (q_1, q_2, \ldots, q_j)$. Let's take the following hypothetical diet matrix, composed of five individuals and four prey categories as an example:

Individual	Food type A	Food type B	Food type C	Food type D
1	8	7	2	3
2	15	3	0	0
3	0	7	3	8
4	0	1	5	10
5	0	0	4	12

The calculation of q_j is straightforward and can be done by summing up all prey items falling into category *j* (sum all *i* individuals) and then converting it into a proportion by dividing it by the total number of prey items of the total population diet:

$$q_j = \frac{\sum_i n_{ij}}{\sum_i \sum_j n_{ij}}$$

In the example, the population diet proportions would be:

Food type A	Food type B	Food type C	Food type D
0.26	0.20	0.16	0.38

so that **p** = (0.26, 0.20, 0.16, 0.38).

4) Prey isotopes

Here you should enter the vector **i** of the empirically determined prey isotopes, in common delta notation. In the previous example, this could be **i** = (-22.28, -21.67, -13.32, -19.39).

5) Prey dry masses

It is at the user's option to incorporate the empirically determined prey dry masses in the simulations. If this option is chosen, you should enter the vector **m** of prey dry masses in any arbitrary units (e.g. mg). For example, **m** = (2.04, 1.58, 0.52, 0.25) could be the vector of dry masses (mg) in the previous example.

6) Number of replicates

The simulations will generate populations with different degrees of among-individual diet variation. Initially populations with extreme degrees of diet variation are generated and diet variation gradually decreases towards zero during the course of simulations (see below). The user must determine how many replicates of simulated populations must be generated at each level of diet variation. Based on our own experience, 100 replicates at each level of diet variation is an appropriate number. This will generate 5,700 simulated populations, which is a fairly good number to establish the relationship between diet and isotope variation and is not computationally prohibitive.

7) Simulations

7.1) Indices of diet variation

We chose two indices of among-individual diet variation. The first is Roughgarden's (1979) *WIC/TNW*, in which the population's total niche width (*TNW*) is partitioned into a within-individual component (*WIC*) and a between-individual component (*BIC*), so that $TNW = WIC + BIC$ (note that Roughgarden referred to these as within- and between-

phenotype components). One can then measure diet variation by calculating the ratio *WIC/TNW*. This index varies from 0 (maximum diet variation) to 1 (no diet variation). While it may be more intuitive to use *BIC/TNW* as a measure of individual specialization because larger values reflect more diet variation, we stick with *WIC/TNW* to follow historical precedent. This index uses the Shannon-Weaver index as an estimate of *TNW* (Roughgarden 1979). As an alternative, the program also calculates a second measure of individual specialization (*IS*) based on distribution-overlap, which assumes the value 1 if there is no diet variation among individuals and tends to 0 as variation increases (Bolnick et al. 2002). Readers are referred to Bolnick et al. (2002) for the formulas of the indices and details on their calculation.

7.2) Generating simulated population

In this section we will briefly discuss how simulated populations are generated. We refer readers to Araújo et al. (manuscript) for details on the simulations.

As previously stated, each simulated population is composed of the empirically observed number of individuals, *N*. Each individual's resource distribution is assigned by a multinomial sample from the empirical population's resource distribution **p**. We can control the level of diet variation among individuals by setting the number of multinomial draws that each individual takes from the population's distribution. Due to the Law of Large Numbers, individuals given few draws have narrower and, as a consequence, more variable diets than when individuals have many draws. Each simulated individual is given *s* random draws (with replacement) from this multinomial probability distribution.

The goal is to use the resulting number of draws (n_{ij}) of each prey type j to represent a long-term diet vector $\mathbf{p}_i$ for the simulated individual. Although we acquire this vector by a sampling process, we use it to represent the vector of individual long-term diet preferences. If an individual is given only a single draw ($s = 1$), it will persistently specialize on a single type of prey resource, e.g., $\mathbf{p_i} = (1.0, 0, 0 \ldots 0)$. Since different individuals will draw different prey from the population vector, $s = 1$ yields the maximum level of among-individual variation. As s increases, individuals' diet vectors $\mathbf{p_i}$ converge towards the population diet vector $\mathbf{p}$ (Law of Large Numbers) and diet variation declines.

After calculating the $\mathbf{p}_i$ vectors, our simulation uses the empirically-obtained prey isotope signatures to calculate each simulated individual's isotope signature:

$$E(\delta_i) = \sum_j p_{ij}\delta_j$$

In case prey dry masses were also entered, our simulation instead uses both the isotope signatures and dry masses to calculate the individuals' isotopic signature:

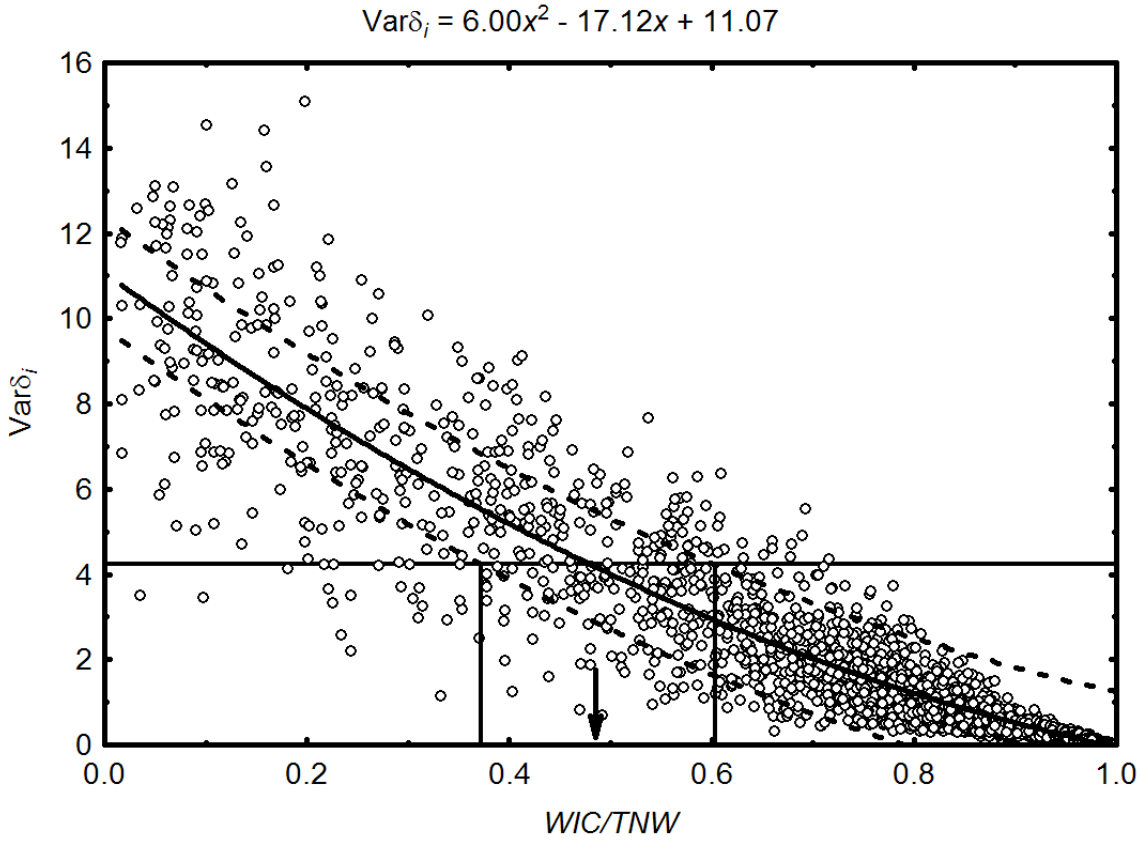
$$E(\delta_i) = \sum_j \frac{p_{ij}m_j}{\sum_j p_{ij}m_j}\delta_j$$

The program then calculates the population isotope variance $\mathrm{Var}\,\delta_i$. Finally, the program calculates *WIC/TNW*, *IS*, and $\mathrm{Var}\,\delta_i$ for the simulated population. The model repeats this procedure for n replicate populations for each of 57 values of s (ranging from 1 to 1,000 in increasing increments).

8) Output

The program outputs a .txt file named VarIso with three columns named 'WIC/TNW', 'IS', and 'Var (isotopes)', in which each line corresponds to one simulated population. This file can be easily imported into Microsoft Excel or common statistics programs to generate scatter plots and regression equations. With this regression equation, one can convert the empirically-estimated isotopic variance into a *WIC/TNW* (or *IS*) value that can be compared with similar measures for other populations. Moreover, regression prediction bands can be estimated and used to establish a confidence interval around the estimated value of *WIC/TNW* (or *IS*). Users can then use the estimated *WIC/TNW* and its confidence interval to check the *WIC/TNW* value estimated from gut contents, as a way of comparing both approaches (see Araújo et al., manuscript). Both *WIC/TNW* and *IS* can be calculated from gut contents with IndSpec1, a program to calculate indices of individual specialization (Bolnick et al. 2002).

To illustrate the use of our method, we provide an example, using the hypothetical sample presented earlier in this manual. Recall that for this sample, **p** = (0.26, 0.20, 0.16, 0.38), **i** = (-22.28, -21.67, -13.32, -19.39), and **m** = (2.04, 1.58, 0.52, 0.25). We also assumed that $N = 30$, and that the empirical $\mathrm{Var}\delta_i = 4.25$. We used STATISTICA7.0 to generate the quadratic regression and prediction bands shown below. We used the resulting equation, and the empirical value of Var δ_i, to solve for an estimated value of *WIC/TNW*.



The solid curve indicates the fitted regressions; the dashed curves are the prediction bands of the regression; the horizontal solid line indicates the empirically estimated $\mathrm{Var}\,\delta_i = 4.25$; the vertical solid lines define the confidence limits around the expected *WIC/TNW* = 0.48, which is indicated by the arrow. The expected *WIC/TNW* was interpolated from the empirical $\mathrm{Var}\,\delta_i$ using the regression equation.

In case an estimate of the variance in fractionation among individuals (Var_Δ) is available for the organism being studied, this can be used to correct the estimate of the empirical $\mathrm{Var}\,\delta_i$ before interpolating the expected *WIC/TNW*. This can be done by simply subtracting Var_Δ from $\mathrm{Var}\,\delta_i$. See Araújo *et al.* (in press) for a detailed discussion on this topic.

9) Troubleshooting

This program is brand new and has not been extensively tested. If you have trouble, please write to either danbolnick@mail.utexas.edu, or msaraujo@gmail.com, giving us as much detail as possible on the problem.

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References

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